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FAX TRANSMITTAL

DATE:	April 8, 2005
TO:	Qian Janice Li
FAX PHONE NO.:	703-872-9306
FROM:	Deborah Martin, Assistant to Sheree Lynn Rybak, Ph.D.
RE:	U.S. Application No. 09/857,719 filed 12/3/2001 For GENE THERAPY FOR CARDIOMYOPATHY
OUR FILE:	6235-59216-01
NO. OF PAGES	3 (including this cover page)

PLEASE ACKNOWLEDGE RECEIPT BY RETURN FACSIMILE? ☐Yes ☒No

CONFIRMATION TO FOLLOW? ☐Yes ☒No

CONTACT INFO: If you do not receive all pages or if you have problems receiving transmittal, please call us at (503) 595-5300 as soon as possible and ask for Deborah Martin.

MESSAGE: Examiner Li--Attached are pages 1 and 11 of the specification, as you requested by telephone on April 7, 2005.

DESCRIPTION

GENE THERAPY FOR CARDIOMYOPATHY

Technical Field

The present invention relates to a method of gene therapy for treating myocardiopathy by noninvasive administration of an HGF (hepatocyte growth factor) gene and therapeutic agents used therefor. More specifically, the present invention relates to a method of gene therapy for treating myocardiopathy by noninvasive administration of an HGF gene into the cardiac muscle, especially to a method of gene therapy that more efficiently treats heart disease, such as cardiomyopathy, angina pectoris and heart failure, by injecting an HGF gene into the affected part of cardiac muscle under the usage of echo, and to therapeutic agents used therefor. Moreover, the present invention relates to a method of gene therapy which is applicable to genes other than HGF genes and that consists of administering genes to the affected part of tissue noninvasively under the usage of echo.

Background Art

In spite of the recent striking technical improvements in the medical field, many problems remain unsolved. The problem of myocardiopathy is one of the important unsolved subjects.

Myocardiopathy is a general name for diseases attributable to organic and functional abnormalities of the cardiac muscle. For example, cardiomyopathy is classified into secondary cardiomyopathy, which occurs in sequence to hypertension, dysbolism, ischemic disease and such, and idiopathic cardiomyopathy (ICM), which occurs without any distinct fundamental disease. Hypertrophic cardiomyopathy (HCM) is classified as an ICM, whose cause of disease is most revealed at the genetic level. In half the numbers of patients with HCM, familial history following autosomal dominant heredity is recognized. Linkage analysis of such family lines, with multiple patients as the object, revealed 5 causal loci so far and the causal gene itself is specified in 4 of them.

Many cases of dilated cardiomyopathy (DCM) occur independently,

(LamatLB9507(BERTHOLO)).

2. Under the usage of echocardiogram (MD500, YOKOKAWA-GE), HVJ-liposome agent was injected into the abdominal lateral cardiac muscle of the heart of myocardiopathy hamster (12 weeks old) and was subjected to following investigations:

1) Density of blood capillary in the cardiac muscle was measured by ALP (alkaline phosphatase) staining and the result of the HGF gene was compared to that of the control.

2) Bloodstream of the heart to which HVJ-liposome was administered was evaluated by laser Doppler imager (LDI) score and the result of the HGF gene was compared to that of the control.

3) After Masson staining of the cardiac muscle, distribution density of fibrosis was measured by computer analysis. Result of the HGF gene was compared to that of the control.

Reference 1

Preparation of HVJ-liposome agent

10 mg Dried lipid (a 1:4.8:2 mixture of phosphatidyl serine, phosphatidyl choline and cholesterol) and 200 µl balanced salt solution (137 µM NaCl, 5.4 µM KCl, 10 µM Tris-HCl; pH7.6) containing HGF gene (100 µg)-HMG1 (high mobility group 1 nuclear protein, 25 µg) was mixed and ,by stirring vigorously with ultrasonication, liposomes were formed. Purified Sendai virus (Z strain) was irradiated with UV (110erg/mm^2/sec) for 3 minutes. Liposome suspension was mixed with Sendai virus (HVJ), heated at 4°C for 10 minutes, and then heated at 37°C for 30 minutes. Free HVJ was discarded and thus obtained HVJ liposome agent.

Reference 2

Measurement on luciferase activity

Liposome agent with 10 µg of luciferase gene was administered to hamsters (6 animals per group). A week later, luciferase activity was measured. Results are shown in Figure 1.

As shown in Figure 1, high levels of luciferase activity were exhibited in the heart. Thus, it was revealed that gene transfer under the usage of echo is possible.